

LARVAL TREMATODES FROM AUSTRALIAN FRESHWATER MOLLUSCS
PART IX

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Cercaria ellisi n. sp.

(Fig. 1-6)

A new echinostome cercaria with 45 collar spines has been studied in the laboratory for several years. It is frequently obtained from *Limnaea lessoni*, from the Murray at Tailcm Bend, and is the only echinostome cercaria, with the exception of *C. Paryphostomi-radiati*, so far noticed by us from this snail host. During the months mentioned the following numbers of snails were found infected with it: May 1937, 10 out of 119 collected; December 1937, 100 of 639; April 1938, 1 of 12; October 1939, 1 of 4; February 1940, 2 of 63; November 1940, 1 of 15; February 1941, 1 of 106; January 1942, 6 of 116; February 1942, 2 of 96; March 1941, 30 of 883; May 1942, 1 of 8; March 1943, 1 of 3—a total of 156 out of 2,064 examined during the period October to May, *i.e.*, 7.5%. It was not recognised in collections made on other occasions during the spring, summer and autumn 1937-1944.

The cercariae are almost incessantly active and exhibit in swimming the typical echinostome figure of 8. When the movement slackens, the tail moves slowly from side to side and the body straightens and is thrust forwards. They are negatively phototropic. The greatest numbers of cercariae are emitted between 11 and 12 in the morning.

The body measurements (in micra) given below are taken from 10 specimens killed by adding to the liquid containing them an equal volume of boiling 10% formalin: length of body from 190-239 μ (average 224); across region of ventral sucker 129-141 μ (average 136); anterior sucker 34-42 μ long (average 38) by 38-46 μ wide (average 42). The posterior sucker was difficult to measure, since in most specimens it was flattened, as in fig. 1. The length in such cases was from 23 to 34 μ , but in well-extended specimens ranged from 46 to 53 μ . The breadth was more constant, varying from 57 to 65 μ (average 61). No satisfactory sucker ratio could be ascertained, though their relative breadths in compressed specimens are about 1:1.5. The distance of the posterior sucker from the anterior end of the cercaria varied from 103 to 148 μ (average 129), and the length of the tail from 342 to 440 μ (average 391). There is no finfold on the tail.

The collar, which is not very evident, bears 45 inconspicuous spines (including four corner spines at each end) arranged in two rows. The spines of the aboral row are slightly longer than those of the oral series. One spine from the aboral row and one from a corner group both measured 11.9 μ . On the ventral and dorsal surfaces minute spinules are arranged regularly as far as the level of the ventral sucker.

The alimentary system is typical of echinostomes. The pharynx is succeeded by a relatively long oesophagus, from which the intestinal caeca arise at the level of the anterior border of the acetabulum and extend to the urinary bladder. Cystogenous cells are numerous and finely granular. The glands were not seen. The genital anlage consisted of two cell masses connected by a string of cells, one mass slightly posterior to the ventral sucker, the other on a level with the anterior border of the ventral sucker.

The excretory system is typical of echinostomes. The cercariae were studied in equal parts of horse serum and water. In most specimens 22 flame cells were seen on each side, arranged as in fig. 1, and appeared to be grouped in threes, but their connections were extremely difficult to work out. These cells opened into a descending ramus which, near the base of the bladder, connected with an ascending ramus. The latter had 15 ciliated patches arranged as in fig. 1, and its convolutions were fairly constant in the cercariae studied. On a level with the top of the pharynx it loops around to enter the main excretory tube. This, as far as the level of the acetabulum, is filled with many small granules, two or three being present in cross section. At the level of the acetabulum the concretions cease, and the main tube forms a characteristic bend in towards the centre and continues to the bladder. The latter is in two parts, a smaller anterior and larger posterior. From the latter a median tube extends for a short distance into the tail and opens by two

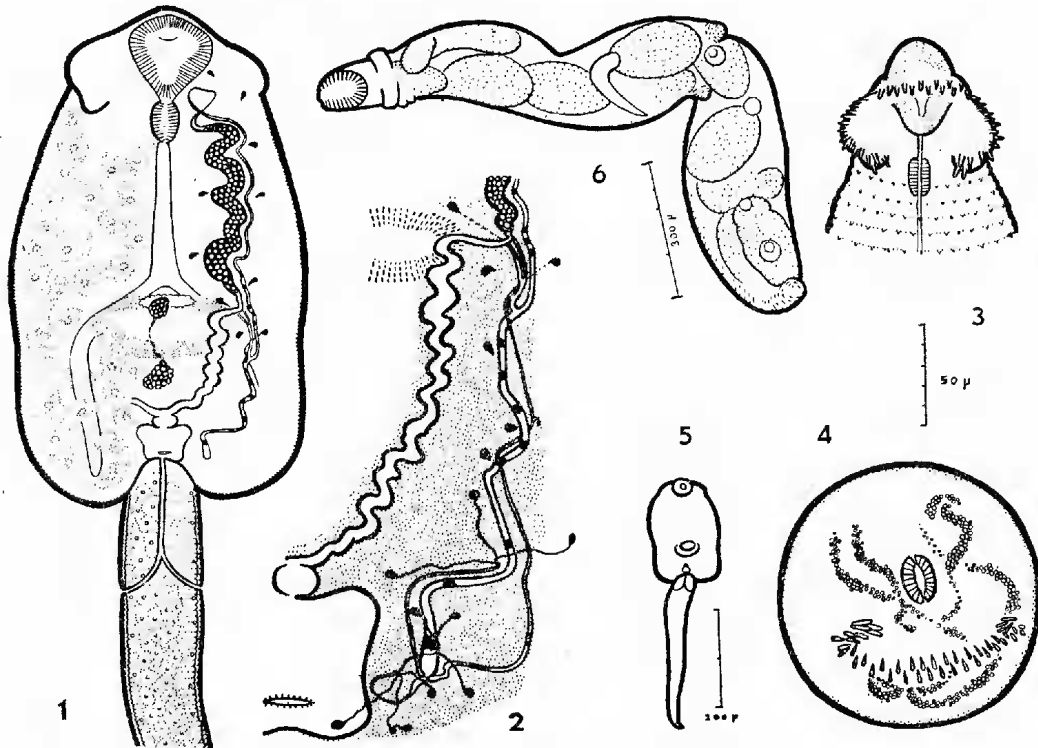


Fig. 1-6—*Cercaria ellisi*: 1, cercaria, collar spines omitted; 2, enlarged freehand diagram of posterior part of excretory system of one side to show position of ciliary flames and flame cells; 3, head end, showing spination; 4, cyst; 5, cercaria, general appearance; 6, redia. Fig. 1, 3 and 4 drawn to scale below fig. 3.

short branches laterally. A small external opening is present on the dorsal surface of the bladder.

Cysts have been obtained experimentally from the following hosts: *Amerianna pyramidata*; *A. tenuistriata* (abundant in mantle cavity); *Planorbis isingi*; *Limnaea lessona*; *Plotiopsis tatei* (abundant in mantle cavity); *Corbiculina angasi* (a few); and a tadpole, *Crinia signifera* (numerous in kidneys). The almost circular cysts varied in measurement between 118 and 133 μ ; 10 from tadpoles averaged 126 by 125 μ , while those from snails were slightly smaller, 122 by 125 μ .

Two young tadpoles were placed in water infested with *C. ellisi*, and within a few minutes cercariae were seen creeping over the surface of the tadpoles and

entering and emerging through the various apertures. After four hours the tadpoles were killed. Twenty-nine cysts and five tailless cercariae were found in the tissues of the mesonephros of one; they were present in least numbers amongst the tubules of the kidney, and in greatest numbers massed near the glomeruli along the nephric ducts. They were also present in the mesenteries.

Another tadpole, killed two hours after having been placed in infected water showed, in addition to the positions mentioned above, two cercariae encysted in the auricle, and several around the heart and aorta, in addition to one in the lung.

Cysts were fed to a canary in February and March 1942, and to a fowl in November and December 1939, but the adult stage was not obtained.

The specific name is given in recognition of assistance received for many years from Mr. L. Ellis, of Tailem Bend and Murray Bridge.

Cercaria ellisi most closely resembles *C. clelandae* Johnston and Angel 1939. When killed under the same conditions our cercaria is slightly shorter and wider than *C. clelandae* (230-290 μ by 89-130 μ). This difference is more marked in the metacercaria, the cysts of *C. clelandae* being consistently 30 μ larger in diameter than those of the present species. *C. clelandae* could not be made to encyst in tadpoles which are a normal secondary host of *C. ellisi*. The rediae in our species are similar but grow to a larger size and contain more developing cercariae. The gut is dark-coloured in *C. clelandae* and inconspicuous in our form. Slight differences occur in the excretory system, which was extremely difficult to work out. Seventeen flames and 24 flame cells have been counted on each side in *C. clelandae*, but only 15 flames and 22 flame cells were seen on each side in our form. As the flame cells are inconspicuous, one or more may have been overlooked.

During January 1943 faecal material deposited by a pelican at Tailem Bend was placed in an aquarium along with several *Limnaea lessoni*, *Amerianna* spp., *Hydrobia* and *Segmentina australis*, and some carp. The snails were tested at the end of February, and weekly after that. Ninety days later two of the *Limnaea* were observed to be giving off a 45-spined echinostome cercaria closely resembling *Cercaria ellisi*, and continued to do so until they died on 5 May 1943 and 5 June 1943 respectively. The remaining snails which had not already died before the latter date, showed no infection.

The body length of these cercariae measured 201 to 243 μ (average 216); the maximum breadth 106-125 μ (average 118); and the tail 293-343 μ long (average 326). The anterior sucker was 42 to 57 μ long (average 46); the posterior sucker 42-57 μ (average 46) long by 46-49 μ (average 48) across, and its distance from the anterior end of the cercaria varied from 106 to 140 μ (average 114). These measurements are similar to those given above for *C. ellisi*, the main differences being in the breadth of the ventral sucker and the length of the tail, which are somewhat less in this form. The material examined consisted of preserved free cercariae as well as of others taken from preserved *Limnaea* snails, and the former may not have been mature when measured. The collar spines are similar to those of *C. ellisi*, and body spines are present on the dorsal and ventral surfaces down to the ventral sucker.

Numerous cysts were found in the liver (particularly at the apex) and mantle cavity and scattered throughout the tissues of the longer-lived host snail. The average measurement of ten cysts was 120 by 121 μ . 114 μ was the lowest and 125 μ the greatest length measurement observed by us. The *Limnaea* snails used in the experiment were laboratory bred and free from infection. In experimental infections of snails, to obtain cysts of *C. ellisi*, it is unusual to find these elsewhere in the body than in the mantle cavity. It is possible that the cysts present in the liver belonged to cercariae which had encysted there instead of emerging

and then encysting in a suitable host. It was very difficult to count the oral spines of these metacercariae, but there seemed to be about 43. We regard this form as belonging to *C. ellisi*.

The characters of the collar spines of *C. ellisi* (and *C. clelandae*) indicate that the adult is probably a species of *Echinostoma* or allied genus. The vertebrate host is probably a bird whose diet includes freshwater molluscs. Nicoll (1914, 112) described *Echinostoma hilliferum*, a 47-spined species from a coot, *Porphyrio melanotus*, North Queensland. *Echinostoma bancrofti* Johnston (1928, 140), described from a waterhen, *Gallinula tenebrosa*, from the Burnett River, Queensland, is recorded to have about 44 collar spines arranged in two alternating rows, with the four corner spines larger and more prominent, but the actual number of oral spines is more likely to be 43 or 45. *Gallinula tenebrosa* and other species of waterhens and coots occur abundantly in the swamps at Tailem Bend. We have not yet found in the pelican an echinostome with 45 collar spines. Waterhens and pelicans frequent the same narrow bank between the swamp and Murray River on which the faecal sample was collected. Contamination of the material from a pelican with that from a waterhen was thus possible.

***Cercaria gigantura* var. *grandior* nov.**

(Fig. 7, 9-11)

On 27 January 1943 one, and on 24 February 1943 two, snails of *Amerianna pyramidata*, from Tailem Bend, were found giving off a cercaria closely resembling *C. gigantura* Johnston and Angel 1941, which these authors regarded as the larva of *Petasisiger australis*. On the latter date two snails also (*Amerianna pyramidata*) gave off *C. gigantura*, and it was possible to study the two cercariae side by side in the laboratory (fig. 8, 9). Macroscopically they appeared to be quite distinct. *C. gigantura* var. *grandior* was much the larger and was relatively a sluggish cercaria. The resting period was usually 4-5 seconds (2-3 seconds in *C. gigantura*); and the tail, because of its greater size, did not move as freely from side to side as that of *C. gigantura*.

Microscopically *C. gigantura* var. *grandior* differed in the following characters: The tail was considerably larger, varying in length from 571 to 1,175 μ , with an average of 717 μ in ten specimens (*C. gigantura* 434 to 584 μ), its breadth ranging from 144 to 245 μ , with an average of 184 μ (*C. gigantura* 134 to 200 μ). The longitudinal muscles were more distinct in our variety, and the circular muscles less so. There was no clear area between the central longitudinal muscle strand and the outer edge (a consistent feature in *C. gigantura*), and the tail was much less transparent. The myomere cells in the tail were also smaller. The tail whip was neither as distinctly marked off, nor as long, as in the typical form, its approximate length being 53 to 114 μ with an average of 79 μ (*C. gigantura* 83-192 μ).

The body of the cercaria seemed in no essential particular other than size to differ from that of *C. gigantura*. The measurements of the variety, with those of *C. gigantura* added in brackets for comparison, were as follows. Length of body 137-300 μ , average 258 μ (105-267); breadth of body across ventral sucker 57-103 μ , average 70 μ (50-100 μ); length of anterior sucker 30-38 μ , average 34 (21-30 μ); breadth of anterior sucker 30-38 μ , average 34 μ ; length of posterior sucker 34-42 μ , average 36 (21-30 μ); breadth of posterior sucker 34-42 μ , average 36 μ (28-38 μ). On remeasurement we found that the size of the ventral sucker of *C. gigantura* fell within the range of that of the variety. The spinules present on the ventral surface of the body were slightly larger in the new variety. The position and character of the collar spines were identical. The length of the latter was 5 μ . Their length in *C. gigantura* was given in error as 13 μ , but on re-

measurement they were found to have the same length ($5\ \mu$) as in the variety; thus bringing the species closer to *C. Petasigeri-nitidi* Beaver 1939.

Our snail hosts died before work on the excretory system of the cercaria was completed. The details found agreed with those of *C. gigantura*. In addition, there was seen in the proximal part of the tail of one specimen a short tube apparently connecting with the excretory bladder in the tail stem. This tube had two short arms, hence it was possible that the excretory tube extended into the

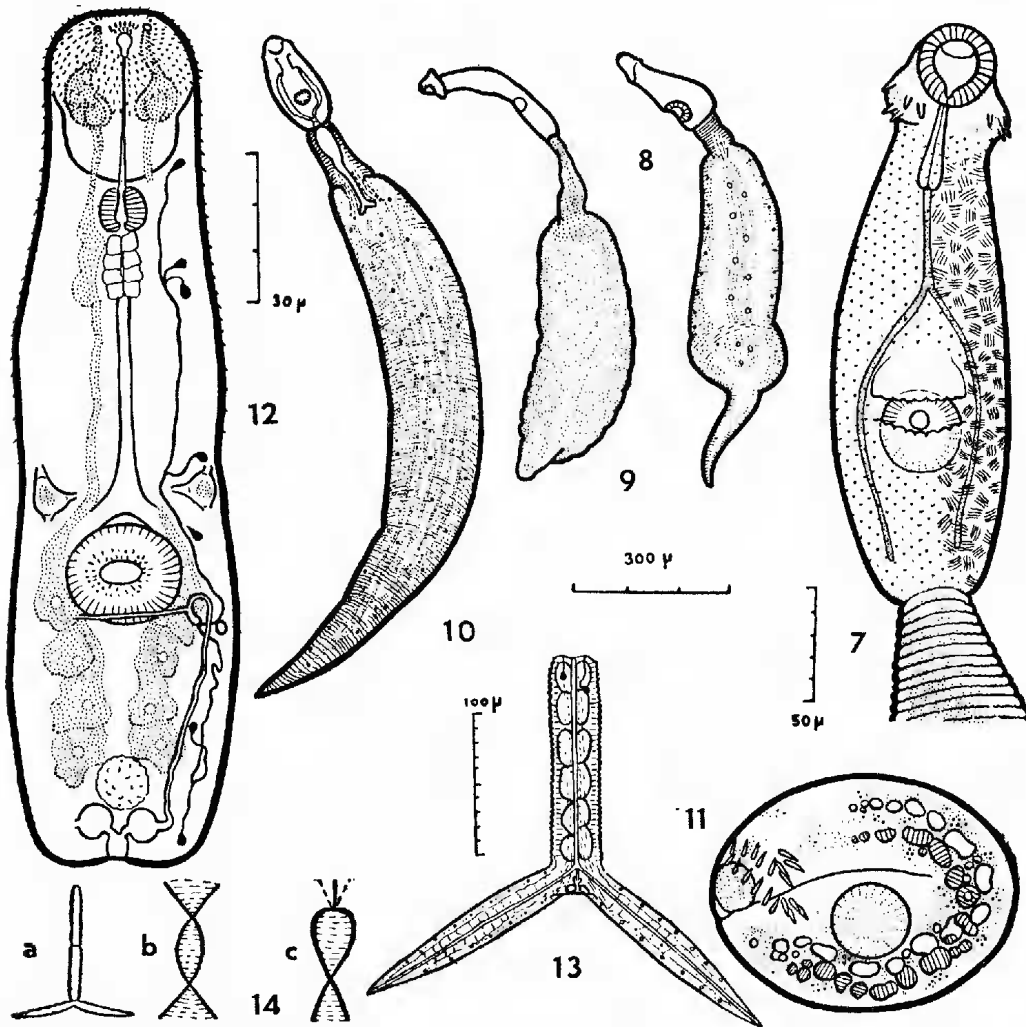


Fig. 7-14. fig. 7, 9-11—*Cercaria gigantura* var. *grandior*: 9, with tail contracted; 10, with tail elongated—its more usual condition while alive; 11, cyst. Fig. 8—*Cercaria gigantura* (typical), for comparison with its variety, fig. 9. Fig. 8-10 to same scale (below 9); 7 and 11 to same scale (beside 7). Fig. 12-14—*Cercaria angelae*: 12, body; 13, tail; 14, a, b, c, freehand sketches of attitudes of living cercariae.

tail for a short distance and opened by two branches laterally. The redia was similar to that of *C. gigantura*.

Cercariae were found experimentally to encyst around the oesophagus and pharynx of the aquarium fish, *Gambusia affinis*, and leopard fish, *Phalloceros caudimaculatus*. Cysts were not recovered from a *Barbus* exposed to infection at the same time. The cysts were similar to those of *C. gigantura* but the dimen-

sions were slightly larger, ranging from 129 to 133 μ in length, and 91 to 99 μ in breadth (*C. gigantura* 125 by 75 μ).

***Cercaria angelae* n. sp.**

(Fig. 12-14)

Cercaria angelae is a small furcocercaria which has been found consistently, though not frequently, emerging from its snail hosts, *Amerianna pyramidata* and *A. tenuistriata*, from the Taillem Bend swamps. The examination of snails for its presence took place from 15 January 1941 to 3 February 1944, and the percentage infection for the period was 0.9%. The largest number of infected snails was observed on 15 January 1941, when 27 out of 861 snails were found giving off the parasite. We have associated with this new form the name of our colleague, Miss L. M. Angel.

C. angelae remains fairly evenly distributed in water, and is an active and vigorous swimmer, appearing to have no resting period. The maximum emission from infected snails occurred between 12 noon and 1.30 p.m., during the summer months. Cercariae collected during these hours were all dead by 9 a.m. the next morning.

The following measurements of 10 cercariae were obtained after adding to the water containing them an equal volume of boiling 10% formalin: Body length 111-216 μ (average 158 μ); body breadth 27-53 μ (average 41); length of tail stem 163-190 μ (average 175); breadth of tail stem 30-46 μ (average 36); length of furcae 163-190 μ (average 176); length of anterior organ 30-46 μ (average 37); length of posterior sucker 19-23 μ (average 19); breadth of posterior sucker 19-23 μ (average 22); distance of posterior sucker from anterior end of cercaria 53-110 μ (average 77).

About eight rows of irregularly-placed spines are present, forming a collar immediately behind the mouth. Two rows of irregularly situated spines (about 12 in number) lie immediately dorsal and anterior to the mouth. Scattered spines, difficult to see, occur on the anterior part of the body, and two irregular rows of spines are present on the ventral sucker. The body has about the same width as the tail stem, and the furcae and tail stem are of almost equal length.

The mouth is subterminal and the pharynx is immediately behind the anterior organ. The latter is more muscular in its posterior portion, though not markedly so. Immediately behind the pharynx are eight large nucleated cells staining deeply with neutral red. The oesophagus is difficult to see and divides into two immediately in front of the ventral sucker. The length of the intestine was not determined owing to the presence of gland cells obscuring it.

Four pairs of large, granular, irregularly-shaped penetration glands, staining with neutral red, are present posterior to the ventral sucker. Their ducts lead anteriorly (fig. 12) with irregular swellings opposite the nucleated post-pharyngeal cells, and with an enlarged section in the anterior part of the anterior organ. A pair of clear cells, with a central granular portion, are present just anterolateral from the ventral sucker. They do not stain with neutral red; perhaps these preacetabular bodies (of Cort and Brackett 1937) are non-pigmented eyes. The reproductive anlage is represented by a group of cells, staining with haematoxylin, in the posterior part of the body. Six well-defined caudal bodies are present in the tail stem. The first caudal body is often considerably smaller than the remainder. A small pair of caudal bodies is present at the base of the furcae. They vary considerably in shape and size, the size probably depending on the age of the cercaria. In this region two globules of similar material are usually present, attached to the central core which supports the caudal bodies.

The bladder consists of two rounded parts each connecting with a short median section which reaches the tail. From the bladder a slightly coiled collect-

ing tube extends to the level of the posterior part of the ventral sucker. Here it forms a loop, from which arises a postacetabular transverse commissure which joins the loop of the excretory tube on the other side. The main duct then divides into two, an anterior and a posterior collecting tubule. Two flame cells open into the posterior collecting tubule, and five flame cells into the anterior collecting tube. Their arrangement is given in fig. 12. Two flame cells are present in the proximal part of the tail. A median excretory tube runs down the tail stem, divides into two branches which open at the tip of the furcae.

The long, much twisted sporocysts, present in the liver of the snail host, are colourless or faintly tinged with yellow, and are hard to disentangle without breaking. One end is pointed and very retractile, the other end rounded, the rest of the body being of uniform thickness except where the cercariae are mature. One sporocyst measured was 2.1 mm. long.

Attempts to obtain the metacercaria by exposing to infection freshwater molluscs, prawns, crayfish and aquatic insect larvae, and fish (*Gambusia*, *Barbus*) and a leech (*Limnodella australis*) were unsuccessful. Many laboratory-bred tadpoles of *Limnodynastes tasmaniensis* were also utilised, most of them unsuccessfully, but in one of them a number of *Tetracotyle* were recovered many months later in the walls of the thorax and rectum, in the pericardium, and in tissues of the tail and those near the base of the forelimbs. The oval cyst, about 325 by 400 μ , had a thick clear outer wall, and a thin pigmented inner wall surrounding the *Tetracotyle* which varied in size in different cysts. The *Tetracotyle* will be described later. The presence of a pharynx was not demonstrated with certainty.

C. angelae is a Strigeid larva belonging to the pharyngeal longifurcate group. The posterior position of the eight gland cells and of the transverse excretory canal, as well as the presence of a well-developed acetabulum suggest that the adult may belong to *Apatemon*, *Strigea*, or perhaps *Apharyngostrigea*.

In swimming constantly during its free life our cercaria is similar to *C. longifurca*, *C. dohema* and the cercaria of *Cotylurus flabelliformis*. Of these three, *C. angelae* in its anatomy somewhat resembles *C. dohema* Cort and Brackett 1937. The measurements of our specimens (which were killed in similar manner to theirs) agree within a few micra with those given for *C. dohema*, except that the furcae appear to be 20 μ shorter in our species. Caudal bodies agree in number and size except that in our specimens the caudal bodies at the base of the furcae are not so large. The caudal excretory tube in *C. angelae* (though not readily seen) opens at the tip of each furca. The excretory bladders are similar; that illustrated in fig. 12 for our species is somewhat contracted, but when it is fully expanded it assumes the form given for *C. dohema*. The number of flame cells is four on each side of the body in *C. dohema*, but there are seven in our species and their arrangement is somewhat different. The nucleated postpharyngeal cells are absent in *C. dohema* which has three instead of four pairs of penetration glands in the postacetabular region.

C. riponi Brackett (1939) from *Stagnicola* from Michigan is another Strigeid cercaria somewhat resembling our form. The most striking differences are in its swimming habits; the different number and arrangement of the flame cells and caudal bodies; the presence of only three pairs of postacetabular gland cells; the smaller preacetabular bodies; and the absence of the postpharyngeal group of nucleated cells.

The presence of four pairs of longitudinally arranged postacetabular glands has been rarely recorded. Lutz (1933, 35, 40, 53, pl. ii, fig. 8) mentioned and figured a "Pseudodistomulum" with such an arrangement. This organism, which is really a cercaria that has shed its tail, was found encysted in a Brazilian frog, *Hyla crepitans*, and a *Tetracotyle* resembling it was reported by him as occurring in young birds, snakes and some carnivores. The adult stage was stated to be

Strigea vaginata (Brandes) from Brazilian Accipitrine birds. Dubois (1938, 94, fig. 37) republished Lutz's figure as *Mesocercaria Strigeae-vaginatae*. From the foregoing it is most probable that the adult of *C. angelae* is a *Strigea* from an Australian bird of prey, several species having already been recorded from hawks and owls from North Queensland.

C. pseudoburti Rankin 1939 has four pairs of postacetabular gland cells, but their arrangement is different from that in *C. angelae*. In *C. burti* Miller the eight postacetabular glands are arranged in two transverse series (Cort and Brooks 1928, pl. xxviii). This cercaria is the larva of an *Apatemon* and closely resembles that of *A. gracilis* as described by Szidat (1929). *Cercaria helvetica* Dubois (1929, 94, pl. iv, fig. 14) from *Limnaea* and *Planorbis* in Switzerland has the eight glands postacetabular and arranged in two close longitudinal rows, and has pre- and postacetabular excretory commissures, but the relative lengths of the prepharynx and oesophagus are different from those of *C. angelae*. Dubois' cercaria also belongs to *Apatemon* (Dubois 1938, 96).

The cercaria of *Apharyngostrigea pipientis*, described by Olivier (1940), has its eight glands almost surrounding the acetabulum, and also differs from *C. angelae* in the relative lengths of the prepharynx and oesophagus and in the form of the tail stem, especially when contracted.

SUMMARY

(1) *Cercaria ellisi* n. sp., a 45-spined echinostome, is described from *Limnaea lessoni*. The cyst stage occurs in the molluscs, *Amerianna* spp., *Planorbis isingi*, *Limnaea lessoni*, *Corbiculina angasi* and *Plotiopsis tatei*; as well as in the tadpole of *Crinia signifera*. The adult probably occurs in a Ralline bird, e.g., a waterhen.

(2) *Cercaria gigantura* var. **grandior** nov. from *Amerianna pyramidata* differs from the type form in the characters of the tail and in having slightly larger cysts. The latter occurs in freshwater fish.

(3) *Cercaria angelae* n. sp. from *Amerianna* spp. is a Strigeid larva with eight longitudinally arranged postacetabular penetration glands. The metacercaria is a *Tetracotyle* occurring in tadpoles. Its adult stage is perhaps a species of *Strigea* parasitic in Australian birds of prey or an *Apharyngostrigea* from herons.

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